



RESEARCH ARTICLE

Unveiling soil-transmitted helminth infections and associated risk factors in rural primary schoolchildren in Malaysia

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ARTICLE HISTORY

Received: 4 April 2024

Revised: 19 June 2024

Accepted: 20 June 2024

Published: 30 September 2024

ABSTRACT

Soil-transmitted helminths (STHs) are known as one of the neglected parasitic diseases, leading to significant health issues and associated complications. This study aims to assess the current prevalence of STH infections and the associated risk factors among rural primary schoolchildren in Malaysia. A cross-sectional study was conducted among 638 schoolchildren (7-11 years old) from 10 rural primary schools in five regions of Malaysia. The overall prevalence of STH infections among schoolchildren was 54.5%, with *T. trichiura* being the predominant STH species (50.9%), followed by *A. lumbricoides* (19.6%) and hookworms (7.4%). The highest prevalence of STH infections was recorded in the schools in Perak (96.6%), followed by Pahang (85.4%), Johor (42.1%) and Sabah (6.2%). At the same time, none of the schoolchildren in Sarawak were infected with STHs. The findings also highlighted that the older age group (10-11 years old) exhibited a higher prevalence of STH infection and *T. trichiura* compared to those aged 7-9 years old ($P = 0.01$) among the schools with a high prevalence of STH infections ($\geq 70\%$). Several variables, such as being female (1.9 [1.2, 3.0]) (Adjusted odd ratio [95% confidence interval]), low household income (30.9 [7.0, 136.5]), using untreated water supply (1.9 [1.1, 3.2]), indiscriminate defaecation (1.9 [1.1, 3.1]), indiscriminate garbage disposal (2.8 [1.3, 6.0]), eating with hands (5.9 [3.4, 10.4]) and experiencing pallor signs (2.3 [1.1, 5.0]), emerged as significant predictors of STH infections in this study population. The present study underscores that in specific rural community areas of Malaysia, STH infections continue to pose health concerns among primary schoolchildren. Hence, to ensure the sustained effectiveness of the measures taken to control STH infections, a collaborative and ongoing effort between various stakeholders is imperative to provide targeted support to rural communities, especially those in areas lacking essential amenities and healthcare services.

Keywords: Malaysia; prevalence; risk factor; rural; soil-transmitted helminth.

INTRODUCTION

Malaysia is a developing country with steady economic progress and a vibrant healthcare sector. Despite these advances, soil-transmitted helminth (STH) infections remain a health concern, particularly among rural populations and indigenous communities. STH infections are caused by parasitic helminths that infect humans and reside in the intestines. These infections are common in developing nations with a significant impact on public health and are considered “the cancers of developing nations”. Parasitic helminths affect more than 1.5 billion people worldwide, especially children in low-income areas with poor socioeconomic backgrounds and lack of access to clean water, sanitation and hygiene (WHO, 2023a).

World Health Organization (2023b) has classified STH infections as neglected tropical diseases (NTDs). These are notably absent from

the global health agenda, resulting in a lack of funding and resources channelled towards resolving the issue despite the current emphasis on Universal Health Coverage. Despite their high prevalence, STH infections are considered NTDs because of three main features namely (1) they are more prevalent in underdeveloped communities than developed communities, (2) they are chronic illnesses and not acute illnesses, and (3) their effects on economic and education burdens are difficult to quantify (Parija *et al.*, 2017). Being infected with STH does not commonly lead to death but will cause chronic illness and extended morbidity, leading to various consequences such as anaemia, blood loss, diarrhoea, low performance in cognitive aspects and malnutrition such as iron deficiency, iron deficiency anaemia, vitamin A deficiency, and stunting (de Gier *et al.*, 2016; Silver *et al.*, 2018; Ulukanligil & Seyrek, 2004). Hence, targeted

measures that can be channelled properly to address STH infections in communities with a high prevalence are of utmost pertinence.

Risk factors for STH infections come from various aspects, such as socioeconomic status (SES), hygiene and community living conditions. STH infections have been commonly reported in many countries or regions with low to middle income, particularly affecting underdeveloped communities (Moncayo *et al.*, 2018). A study by Molla and Mamo (2018) highlighted behavioural factors that increase the risk of having STH infections, including the frequency of sucking fingers or nails. Meanwhile, another study reported that sources of drinking water, habits of walking barefoot, and defecation places are also risk factors for acquiring STH infections among children (Eyayu *et al.*, 2022). Based on a study conducted in Southern India (Ajampur *et al.*, 2021), improved sanitation and proper hygiene facilities significantly reduce the risk of STH infections.

Although there are numerous studies conducted on the prevalence of STH infections in Malaysia (Ahmed *et al.*, 2011; Al-Delaimy *et al.*, 2014; Rajoo *et al.*, 2017; Wong *et al.*, 2016), there are still limited studies evaluating their prevalence in rural primary schoolchildren. This scarcity of information warrants an urgent update. Furthermore, previous studies have focused on specific communities or localities with small sample sizes. Hence, within this context, this study aimed to provide comprehensive nationwide data on the current prevalence of STH infections and the associated risk factors among rural primary schoolchildren across five regions in Malaysia.

MATERIALS AND METHODS

Study area, design and populations

A cross-sectional study was conducted from April 2017 to October 2017 in five regions of Malaysia, which included a northern region (Perak state), a central region (Pahang state), a southern region (Johor state), and Borneo (Sabah and Sarawak states). This study serves as a screening phase of a nationwide clinical trial to provide nationwide baseline data on the current prevalence of STH infections and the associated risk factors among rural schoolchildren in Malaysia. The selection criteria for schools and locations and schools were previously delineated (Tan *et al.*, 2022). After random

selection, ten rural primary schools were included in the study, and two schools were selected in each region (Figure 1). Briefly, the study population consisted of students from diverse ethnic backgrounds, including Orang Asli (OA) students predominantly belonging to the Semai and Temiar subgroups, as well as students from Malay, Kadazandusun, and Iban ethnicities (Tan *et al.*, 2022). OA refers to the original or indigenous people, a minority group in Peninsular Malaysia (International Work Group for Indigenous Affairs, 2021).

Ethics approval and consent to participate

The study's protocol received approval from the Medical Research and Ethics Committee, Ministry of Health, Malaysia, under registration number NMRR-16-1905-32547 and was also registered at ClinicalTrials.gov under the identifier NCT03256123. Permissions to conduct the study were also sought from the Ministry of Education, Malaysia and the Department of Orang Asli Development, Malaysia. This study was carried out following the principles of the Declaration of Helsinki. Parents and guardians were duly informed of the voluntary nature of their child's participation and were made aware of their right to withdraw from the study at their discretion. Written informed consent was obtained from literate parents and guardians prior to the commencement of the study. As for illiterate parents and guardians' verbal consent was obtained with their thumbprints on the informed consent forms. A brief explanation was given after the acquisition of consent from the parents and guardians. An assent form containing colourful illustrations and graphics depicting the study's overall information was also obtained from the schoolchildren to ensure that they understood the aim of their participation and that their participation was voluntary.

Sample size estimation

The sample size required for this study was determined based on the expected and most recent prevalence of STH infections documented in similar communities using the convenience random sampling technique. Based on the community-based study conducted in Sarawak, the overall prevalence of STH infections stood at 50.4% (Rajoo *et al.*, 2017). By utilising the following formula developed by Leedy (1993):

$$n \geq (z/m)^2 \times p(1-p)$$

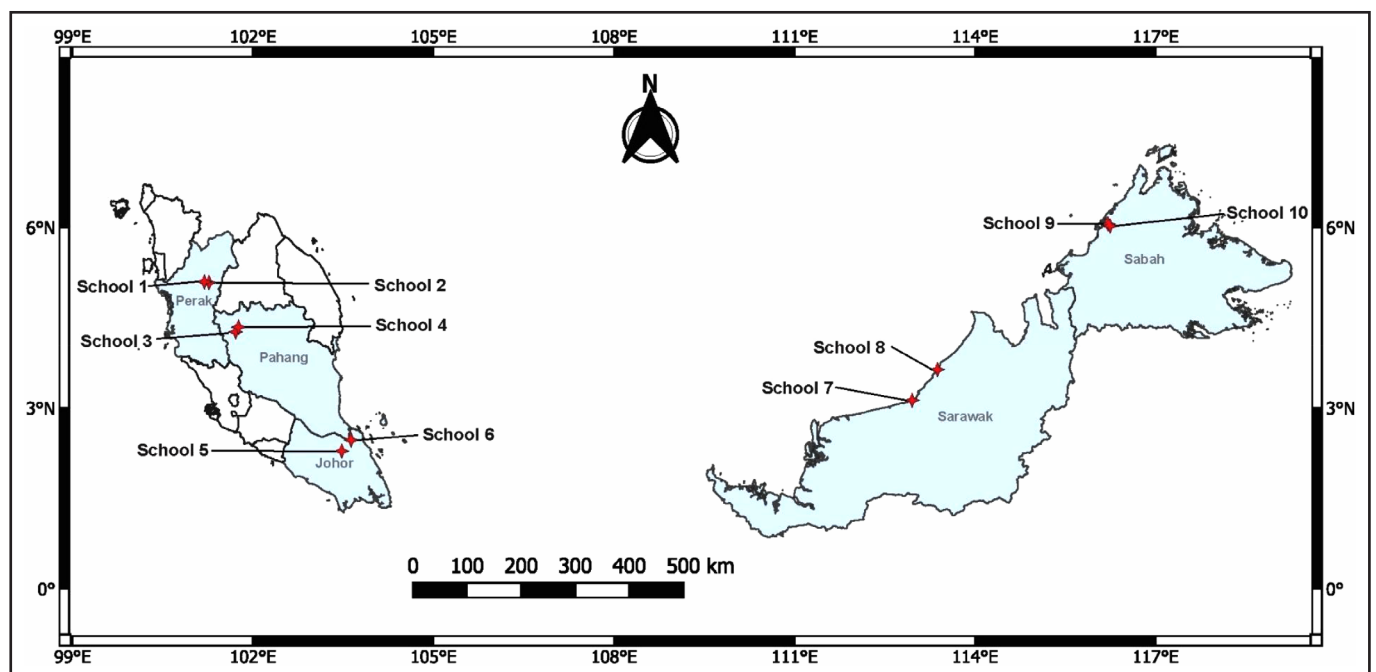


Figure 1. Map showing the locations of the selected schools. A total of ten national rural primary schools were selected from five different regions in Malaysia, including the northern region (Perak state), the central region (Pahang state), the southern region (Johor state), and the Borneo (Sabah and Sarawak states).

The minimum sample size (n) was calculated using the following variables considered: a standard score (z) of 1.96, a sampling error (m) of 5%, and an estimated prevalence or rate of the variable within the population. A minimum sample size of 384 was necessitated in this study, with a 5% significance level and a 95% confidence level. Considering the mixed populations in this study, along with other factors such as the probability of getting incomplete samples and having greater precision on data collected, more schoolchildren were approached according to availability and obtainment of consent from their guardians or parents.

Pre-tested questionnaire

The schoolchildren were instructed to answer a set of pre-tested questionnaires face-to-face by trained fieldworkers with the assistance of teachers from the respective schools. The questionnaires were administered to collect information on various aspects, including sociodemographic data (*i.e.*, age, sex, level of primary education, date of birth and monthly household income), living conditions and environmental sanitation characteristics (*i.e.*, water supply type, latrine system, garbage disposal method and the presence of domestic animals), hygiene behaviours (*i.e.*, wearing shoes, handwashing practices and taking a bath once a day) and health status (*i.e.*, the usage of anthelmintic drugs and iron supplements).

The questionnaires were written in Malay, the national language of Malaysia, and were easily comprehensible to the participants. They were designed with many pictures and colours to help the schoolchildren understand and comprehend the questions more easily. For certain sections of the questionnaires (*e.g.*, monthly household income, date of birth, medical treatment/status), the data was acquired from the student database obtained from the school's administration and the parents.

Stool sample collection and analysis

A plastic container with a wide-mouth screw cap, pre-labelled with the code and name of the participants, was distributed to each schoolchild. The schoolchildren were instructed to scoop their stool samples according to the marked levels. Trained fieldworkers gave a brief demonstration with an infographic and pictures of the sample collection procedure. The schoolchildren were reminded to avoid any urine contamination with the stool samples. The stool samples were then collected on the following day. The collected stool samples were preserved with 2.5% potassium dichromate. Subsequently, these samples were placed in a suitable collection box and transported to the Department of Parasitology, Faculty of Medicine, Universiti Malaya.

The collected stool samples were examined to detect STHs through the formalin-ether concentration technique (Allen & Ridley, 1970). Briefly, a sample was mixed with 10% formalin (7 mL) in a Falcon tube and filtered through gauze cloth into another Falcon tube. Following this step, 2–3 mL of ether was added to the supernatant, and centrifugation at 2,000 revolutions per minute (rpm for 2–3 minutes) was conducted. This centrifugal process led to four distinct layers from top to bottom: ether, a fat or debris plug, formalin, and a pellet. An applicator stick was gently pressed around the tube's circumference to dislodge the fat or debris plug between ether and formalin at the interface. The top three layers were discarded with a single smooth inversion of the centrifuge tube, leaving only the final sediment. Then, the supernatant was discarded by inverting the Falcon tube. A Pasteur pipette was used to withdraw the bottom sediment, which was then mixed with iodine and smeared onto the microscope slides. The slides underwent examination using a light microscope (Olympus, CX40, USA) at $\times 10$ and $\times 40$ magnifications. A sample was deemed STH-positive when at least one STH egg was detected during the microscopy examination.

Statistical analyses

Laboratory analysis outcomes and questionnaire findings were input and analysed using the Statistical Package for the Social Sciences (IBM SPSS Statistics) software for Windows version 22. Before conducting the analyses, the entered data were cross-verified regularly to ensure that all the data were input correctly. For categorical data, frequency and rate (percentage) were used and assessed by the chi-square test (χ^2) and Fisher's exact test. A univariate logistic regression analysis was conducted to test the associated risk factors of STH infections. For each statistically significant factor, an odds ratio (OR) and a 95% confidence interval (95% CI) were used to examine the strength of the association concerning the variable of interest. The variables with a borderline significance level of ≤ 0.25 were incorporated into the multivariate logistic regression analysis and displayed as adjusted ORs alongside their corresponding 95% CI to ensure all potential predictors were included. A significance threshold of $P < 0.05$ denoted statistical significance.

RESULTS

A total of 638 schoolchildren were included in the final analysis (Table 1). The schoolchildren's ages ranged from 7 to 11 during the study period, with both mode and median age of 9. There were 50.9% males and 49.1% females, whereby 57.8% were in lower primary grade (7 to 9 years old) and 42.2% were in upper primary grade (10 to 11 years old). Over half of the study population were OA schoolchildren (58.8%). Most schoolchildren hail from low-income families (92.5%) and have a monthly household income of less than RM500 or approximately USD118. Concerning the geographical distribution, the schoolchildren were predominantly found in Peninsular Malaysia, constituting 66.0%. The central region, Pahang State, had the highest number of schoolchildren at 31.0%, followed by the northern region, Perak State (66.0%) and the southern region, Johor State (11.6%). On the other hand, 34% of the study population originated from Borneo Malaysia, with 21.3% from Sarawak state and 12.7% from Sabah state.

Table 1. General characteristics of the overall schoolchildren population (N=638)

Variables	n (%)
Socio-demographic characteristics	
Age	
7–9 years	369 (57.8)
10–11 years	269 (42.2)
Sex	
Male	325 (50.9)
Female	313 (49.1)
Monthly household income (N = 636) [#]	
<RM 500	588 (92.5)
>RM 500	48 (7.5)
Regions	
Peninsular Malaysia	421 (66.0)
Perak (Northern)	(147). (23.0)
School 1	82 (12.9)
School 2	65 (10.2)
Pahang (Central)	198 (31.0)
School 3	100 (15.7)
School 4	98 (15.4)
Johor (Southern)	76 (11.9)
School 5	46 (7.2)
School 6	30 (4.7)
Borneo Malaysia	217 (34.0)
Sarawak	136 (21.3)
School 7	93 (14.6)
School 8	43 (6.7)
Sabah	81 (12.7)
School 9	39 (6.1)
School 10	42 (6.6)

Type of schools	
Orang Asli* schools	375 (58.8)
Non-Orang Asli schools	263 (41.2)
Living conditions and environmental sanitation	
Source of water supply (N = 633) [#]	
Untreated (river, well, rain, etc.)	170 (26.9)
Treated (government pipe water system)	463 (73.1)
Presence of toilet (N = 634) [#]	
No	114 (18.0)
Yes	520 (82.0)
Toilet facility (N = 530) [#]	
No flush	241 (45.5)
Flush	289 (54.5)
Defaecation sites (N = 633) [#]	
Improper (open/indiscriminate)	382 (60.3)
Proper (toilet/latrine)	251 (39.7)
Garbage disposal (N = 633) [#]	
Improper (indiscriminate)	93 (14.7)
Proper (systematic/collected)	540 (85.3)
Presence of domestic animal (N = 634) [#]	
Yes	603 (95.1)
No	31 (4.9)
Close contact with the animal (N = 633) [#]	
Yes	529 (83.6)
No	104 (16.4)
Hygiene behaviours	
Eating with hands (N = 634) [#]	
Yes	499 (78.7)
No	135 (21.3)
Taking a bath at least once a day (N = 634) [#]	
No	32 (5.0)
Yes	602 (95.0)
Changing clothes at least once a day (N = 634) [#]	
No	18 (2.8)
Yes	616 (97.2)
Wearing shoes outside the house (N = 634) [#]	
No	91 (14.4)
Yes	543 (85.6)
Washing hands before eating (N = 634) [#]	
No	53 (8.4)
Yes	581 (91.6)
Washing hands after defaecation (N = 634) [#]	
No	68 (10.7)
Yes	566 (89.3)
Health status	
Taking an iron supplement (N = 634) [#]	
No	550 (86.8)
Yes	84 (13.2)
Taking anthelmintic drugs (N = 634) [#]	
No	273 (43.1)
Yes	361 (56.9)
Experiencing black stool (N = 634) [#]	
Yes	407 (64.2)
No	227 (35.8)
Experiencing tiredness (N = 633) [#]	
Yes	257 (40.6)
No	376 (59.4)
Experiencing pallor signs (N = 634) [#]	
Yes	68 (10.7)
No	566 (89.3)
Experiencing angular cheilitis (N = 634) [#]	
Yes	31 (4.9)
No	603 (98.5)

* Indigenous minority peoples of Peninsular Malaysia.

[#] Consists of missing values.

Concerning the living conditions and environmental sanitation characteristics, most of the study population had access to a treated water supply (73.1%) and a toilet at home (82.0%). Nevertheless, nearly half lacked a flush toilet facility in their homes, and many were reported to engage in improper defecation practices (60.6%). Most of the study population adhered to proper garbage disposal methods (85.3%). Almost all the participants owned domestic animals (95.1%), with a large percentage having close contact with the animals (83.6%).

As for hygiene behaviours and health status, a significant proportion of the schoolchildren were found to eat with their hands (78.7%), whereas most of the students reported the practice of handwashing before eating (91.6%). Besides, many of the study participants reported that they practise good hygiene behaviours such as taking a bath at least once a day (95.0%), changing clothes at least once a day (97.2%), wearing shoes outside the house (85.6%) and wash hands after defaecation (89.3%). Besides, most of the study population did not take iron supplements (86.6%) and experienced black stool (64.2%). Nearly half the study population did not take anthelmintic drugs within the past 6 months (43.1%) and experienced tiredness (40.6%). In contrast, only a small percentage experienced pallor signs (10.7%) and angular cheilitis (4.9%).

Soil-transmitted helminth (STH) infections status

The prevalence of STH infections among schoolchildren was 54.5%, with *T. trichiura* (50.9%) being the most found species, followed by *A. lumbricoides* and hookworm at 19.6% and 7.4%, respectively (Table 2). Among those who were infected with STH, single infection (63.2%) was the most common, followed by dual infection (30.7%) and triple infection (6.0%). The coinfection of ascariasis and trichuriasis (13.2%) was the most prevalent multiple infections, followed by 3.4% of the children infected with both *T. trichiura* and hookworm, and 3.3% infected with all three types of STHs (*A. lumbricoides*, *T. trichiura* and hookworm).

Based on stratification by schools, the highest prevalence of STH infections was evident in both the schools in Perak state (96.6%): school 1 (96.3%) and school 2 (96.9%). The schools followed this in Pahang state (overall: 85.4%, school 3: 86.0% and school 4: 84.7%), the schools in Johor state (overall: 42.1%, school 5: 19.6% and school 6: 76.7%) and the schools in Sabah state (overall: 6.2%, school 9: 7.7% and school 10: 4.8%). In contrast, none of the schoolchildren in Sarawak state was found positive for any STH.

Subsequently, the schools with a high prevalence of STH infection ($\geq 70\%$), namely schools 1, 2, 3, 4 and 6, were selected for a detailed investigation into the differences in STH infection status among the schoolchildren in these schools after stratification by age group and sex (Table 3). A higher prevalence of STH infection was observed among schoolchildren aged 10-11 years old (93.8%) compared to those aged 7-9 years old (85.6%) ($P = 0.01$). Furthermore, schoolchildren aged 10-11 years old (90.6%) exhibited a higher prevalence of *T. trichiura* in comparison to their younger counterparts (80.5%) ($P = 0.01$). On the other hand, no significant differences were observed between sexes in schools with a high prevalence of STH infection.

Risk factors of soil-transmitted helminth (STH) infections

The risk factors associated with any STH infections among the overall study population, schools with high prevalence of STH infections ($\geq 70\%$) and schools with low prevalence of STH infections ($< 70\%$) are tabulated in Table 4. Based on univariate logistic regression analysis, a number of variables were found to be significantly associated with STH infections, including low monthly household income (OR = 32.7; 95% CI = 7.9–135.8; $P < 0.001$), untreated water supply (OR = 3.7; 95% CI = 2.5–5.5; $P < 0.001$), absence of toilet at home (OR = 17.8; 95% CI = 8.1–39.1; $P < 0.001$), absence of flush toilet (OR = 1.5; 95% CI = 1.1–2.0; $P = 0.02$) indiscriminate defaecation (OR = 2.5; 95% CI = 1.8–3.5; $P < 0.001$), indiscriminate

Table 2. STH infection status of the schoolchildren stratified by schools (N=638)

Characteristics	Overall (N = 638)	Perak			Pahang			Johor			Sarawak			Sabah		p-value
		School 1 (n=82)	School 2 (n=65)	School 3 (n=100)	School 4 (n=98)	School 5 (n=46)	School 6 (n=30)	School 7 (n=93)	School 8 (n=43)	School 9 (n=39)	School 10 (n=42)					
STH infection status*																
Positive	348 (54.5)	79 (96.3)	63 (96.9)	86 (86.0)	83 (84.7)	9 (19.6)	23 (76.7)	0 (0.0)	0 (0.0)	3 (7.7)	2 (4.8)	< 0.001				
Negative	290 (45.5)	3 (3.7)	2 (3.1)	14 (14.0)	15 (15.3)	37 (80.4)	7 (23.3)	93 (100.0)	43 (100.0)	36 (92.3)	40 (95.2)	< 0.001				
Species																
<i>A. lumbricoides</i>	125 (19.6)	46 (56.1)	25 (38.5)	29 (29.0)	7 (7.1)	8 (17.4)	9 (30.0)	0 (0.0)	0 (0.0)	1 (2.6)	0 (0.0)	< 0.001				
<i>T. trichiura</i>	325 (50.9)	77 (93.9)	58 (89.2)	81 (81.0)	83 (84.7)	6 (13.0)	19 (63.3)	0 (0.0)	0 (0.0)	1 (2.6)	0 (0.0)	< 0.001				
Hookworms	47 (7.4)	18 (22.0)	13 (20.0)	4 (4.0)	5 (5.1)	0 (0.0)	4 (13.3)	0 (0.0)	0 (0.0)	1 (2.6)	2 (4.8)	< 0.001				
Type of parasitism																
Single infection	220 (34.5)	31 (37.8)	37 (56.9)	58 (58.0)	71 (72.4)	4 (8.7)	14 (46.6)	0 (0.0)	0 (0.0)	3 (7.8)	2 (4.8)	< 0.001				
Only <i>A. lumbricoides</i>	19 (3.0)	2 (2.4)	5 (7.7)	5 (5.0)	0 (0.0)	3 (6.5)	3 (10.0)	0 (0.0)	0 (0.0)	1 (2.6)	0 (0.0)	< 0.001				
Only <i>T. trichiura</i>	197 (30.9)	29 (35.4)	32 (49.2)	53 (53.0)	71 (72.4)	1 (2.2)	10 (33.3)	0 (0.0)	0 (0.0)	1 (2.6)	0 (0.0)	< 0.001				
Only hookworm	4 (0.6)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	1 (3.3)	0 (0.0)	0 (0.0)	1 (2.6)	2 (4.8)	< 0.001				
Dual infection	107 (16.8)	34 (41.5)	19 (29.2)	28 (28.0)	12 (12.2)	5 (10.9)	9 (30.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	< 0.001				
<i>T. trichiura</i> + <i>A. lumbricoides</i>	85 (13.3)	30 (36.6)	13 (20.0)	24 (24.0)	7 (7.1)	5 (10.9)	6 (20.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	< 0.001				
<i>T. trichiura</i> + hookworm	22 (3.4)	4 (4.9)	6 (9.2)	4 (4.0)	5 (5.1)	0 (0.0)	3 (10.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	< 0.001				
Triple infection (<i>T. trichiura</i> + <i>A. lumbricoides</i> + hookworm)	21 (3.3)	14 (17.1)	7 (10.8)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	< 0.001				

All values are given as n (%).

* Significant difference between type of schools: *p < 0.05; **p < 0.001 (based on Fisher's exact test).

Children infected with any forms of STH (*A. lumbricoides*, *T. trichiura*, and hookworms).

Table 3. STH infection status according to age group and sex among the schools with a high prevalence of STH infections ($\geq 70\%$) (N=375)

Characteristics	Age group		p-value	Sex		p-value
	7-9 years old (n = 215)	10-11 years old (n = 160)		Male (n = 180)	Female (n = 195)	
STH infection status[#]						
Positive	184 (85.6)	150 (93.8)	0.01*	163 (90.6)	171 (87.7)	NS
Negative	31 (14.4)	10 (6.3)		17 (9.4)	24 (12.3)	
Species						
<i>A. lumbricoides</i>	66 (30.7)	50 (31.3)	NS	63 (35.0)	53 (27.2)	NS
<i>T. trichiura</i>	173 (80.5)	145 (90.6)	0.01*	153 (85.0)	165 (84.6)	NS
Hookworms	20 (9.3)	24 (15.0)	NS	20 (11.1)	24 (12.3)	NS
Type of parasitism						
Single infection	117 (54.4)	94 (58.8)	NS	98 (54.4)	113 (57.9)	NS
Only <i>A. lumbricoides</i>	11 (5.1)	4 (2.5)		9 (5.0)	6 (3.1)	
Only <i>T. trichiura</i>	106 (49.3)	89 (55.6)		88 (48.9)	107 (54.9)	
Only hookworm	0 (0.0)	1 (0.6)		1 (0.6)	0 (0.0)	
Dual infection	59 (27.4)	43 (26.9)	NS	57 (31.7)	45 (23.1)	NS
<i>T. trichiura</i> + <i>A. lumbricoides</i>	47 (21.9)	33 (20.6)		46 (25.6)	33 (16.9)	
<i>T. trichiura</i> + hookworm	12 (5.6)	10 (6.3)		11 (6.1)	11 (5.6)	
Triple infection (<i>T. trichiura</i> + <i>A. lumbricoides</i> + hookworm)	8 (3.7)	13 (8.1)	NS	8 (4.4)	13 (6.7)	NS

NS: Not significant.

All values are given as n (%).

* Significant difference between type of schools: * $p < 0.05$; ** $p < 0.001$ (based on chi-square test).

[#] Children infected with any forms of STH (*A. lumbricoides*, *T. trichiura*, and hookworms).

Table 4. Univariate and multivariate analysis of risk factors associated with STH infections among schoolchildren (N=638)

Variables	N	n	%	OR (95% CI)	p-value	Adjusted OR (95% CI)	p-value
Overall population (N = 638)							
Age 10–11 years old	269	154	57.2	1.2 (0.9, 1.7)	0.24	1.2 (0.8, 1.9)	NS
Female	313	180	57.7	1.3 (0.9, 1.7)	0.14	1.9 (1.2, 3.0)	0.003*
Low household income	588	345	58.7	32.7 (7.9, 135.8)	<0.001**	30.9 (7.0, 136.5)	<0.001**
Untreated water supply	170	130	76.5	3.7 (2.5, 5.5)	<0.001**	1.9 (1.1, 3.2)	0.03*
Absence of toilet at home	114	107	93.9	17.8 (8.1, 39.1)	<0.001**	2.4 (0.4, 14.0)	NS
Absence of flush toilet	344	203	59.0	1.5 (1.1, 2.0)	0.02*	0.8 (0.5, 1.4)	NS
Indiscriminate defaecation	382	243	63.6	2.5 (1.8, 3.5)	<0.001**	1.9 (1.1, 3.1)	0.01*
Indiscriminate garbage disposal	93	79	84.9	5.8 (3.2, 10.4)	<0.001**	2.8 (1.3, 6.0)	0.01*
Presence of domestic animal	603	336	55.7	2.3 (1.1, 4.9)	0.03*	1.4 (0.5, 4.0)	NS
Eating with hands	499	317	63.5	6.1 (3.9, 9.5)	<0.001**	5.9 (3.4, 10.4)	<0.001**
Not washing hands after playing with the soil	53	39	73.6	2.5 (1.3, 4.6)	0.004*	1.6 (0.5, 4.7)	NS
Not washing hands after defaecation	68	45	66.2	1.7 (1.0, 2.9)	0.045*	1.2 (0.5, 2.7)	NS
Experiencing tiredness	257	160	62.3	1.7 (1.2, 2.3)	0.002*	1.22 (0.7, 1.9)	NS
Experiencing pallor signs	68	52	76.5	3.0 (1.7, 5.4)	<0.001**	2.3 (1.1, 5.0)	0.03*
Schools with high prevalence of STH infections (≥ 70%) (N = 375)							
Age 10–11 years old	160	150	93.8	2.5 (1.2, 5.3)	0.01*	3.3 (1.4, 7.9)	0.01*
Absence of flush toilet	203	192	94.6	3.5 (1.2, 6.6)	0.02*	2.6 (1.0, 6.5)	0.046*
Indiscriminate defaecation site	249	231	92.8	2.7 (1.4, 5.2)	0.003*	1.6 (0.7, 3.4)	NS
Presence of animals at home	358	322	89.9	2.4 (0.7, 9.2)	0.17	2.3 (0.9, 6.0)	NS
Schools with low prevalence of STH infections (< 70%) (N = 263)							
Female	118	9	7.6	2.3 (0.8, 7.1)	0.13	9.6 (1.4, 65.7)	0.02*
Untreated water source [#]	33	4	12.1	3.0 (0.9, 10.3)	0.08	1.8 (0.3, 10.6)	NS
Absence of toilet [#]	9	6	66.7	61.3 (12.9, 289.9)	<0.001**	287.6 (18.4, 4487.3)	<0.001**
Indiscriminate defaecation site	133	12	9.0	6.3 (1.4, 28.7)	0.01*	2.4 (0.4, 13.2)	NS
Indiscriminate garbage disposal [#]	9	2	22.2	5.7 (1.1, 30.6)	0.08	0.2 (0.01, 3.2)	NS
Not wearing shoes [#]	46	6	13.0	3.9 (1.3, 11.8)	0.02*	10.5 (2.2, 50.2)	0.003*

CI: confidence interval; NS: not significant; OR: odds ratio.

* Significant difference based on univariate and multivariate analysis: * $p < 0.05$; ** $p < 0.001$.

[#] Variable with p-value was calculated based on Fisher's exact test due to the small number of samples in the cells.

garbage disposal (OR = 5.8; 95% CI = 3.2–10.4; $P < 0.001$), presence of domestic animal (OR = 2.3; 95% CI = 1.1–4.9; $P = 0.03$), eating with hands (OR = 6.1; 95% CI = 3.9–9.5; $P < 0.001$), not washing hands after playing with the soil (OR = 2.5; 95% CI = 1.3–4.6; $P = 0.004$), not washing hands after defaecation (OR = 1.7; 95% CI = 1.0–2.9; $P = 0.045$), experiencing tiredness (OR = 1.7; 95% CI = 1.2–2.3; $P = 0.002$) and experiencing pallor signs (OR = 3.0; 95% CI = 1.7–5.4; $P < 0.001$). The final multivariate logistic regression analysis indicated that factors such as being female (OR = 1.9; 95% CI = 1.2–3.0; $P = 0.003$), low household income (OR = 30.9; 95% CI = 7.0–136.5; $P < 0.001$), untreated water supply (OR = 1.9; 95% CI = 1.1–3.2; $P = 0.03$), indiscriminate defaecation (OR = 1.9; 95% CI = 1.1–3.1; $P = 0.01$), indiscriminate garbage disposal (OR = 2.8; 95% CI = 1.3–6.0; $P = 0.01$), eating with hands (OR = 5.9; 95% CI = 3.4–10.4; $P < 0.001$) and experiencing pallor signs (OR = 2.3; 95% CI = 1.1–5.0; $P = 0.03$) were significant predictors of STH infections in the current population.

Among the schools with high prevalence of STH infections ($\geq 70\%$), older age group (OR = 2.5; 95% CI = 1.2–5.3; $P = 0.01$), absence of flush toilet (OR = 3.5; 95% CI = 1.2–6.6; $P = 0.02$), and indiscriminate defaecation site (OR = 2.7; 95% CI = 1.4–5.2; $P = 0.003$) were significantly associated with STH infections. Following that, multivariate logistic regression analysis showed that the older age group (OR = 3.3; 95% CI = 1.4–7.9; $P = 0.01$) and absence of a flush toilet (OR = 2.6; 95% CI = 1.0–6.5; $P = 0.046$) were the significant predictors of STH infections.

For the schools with low prevalence of STH infections ($< 70\%$), the univariate logistic regression analysis showed that the absence of toilet (OR = 61.3; 95% CI = 12.9–289.9; $P < 0.001$), indiscriminate defaecation site (OR = 6.3; 95% CI = 1.4–28.7; $P = 0.01$) and did not wear shoes (OR = 3.9; 95% CI = 1.3–11.8; $P = 0.02$) were the significant risk factors of STH infections. Subsequently, the results of multivariate logistic regression analysis ruled out that being female (OR = 9.6; 95% CI = 1.4–65.7; $P = 0.02$), absence of a toilet (OR = 287.6; 95% CI = 18.4–4487.3; $P < 0.001$) and did not wear shoes (OR = 10.5; 95% CI = 2.2–50.2; $P = 0.003$) were the significant predictors of any STH infections.

DISCUSSION

The present study depicts a high prevalence of STH infections (54.6%) among primary schoolchildren residing in the rural communities of Malaysia. The prevalence of STH infections in this study is slightly lower than both recent and similar studies conducted primarily in Peninsular Malaysia, which ranged from 59.9–93.7% (Ahmed et al., 2012a; Mohd-Shaharuddin et al., 2021; Muslim et al., 2021; Nasr et al., 2020; Ngui et al., 2011; 2012). However, there is a similar prevalence finding in a study carried out among indigenous people in Sarawak, whereby the overall prevalence of STH infection is 50.4% (Rajoo et al., 2017). Most of the previously reported local studies on STH infections were conducted among children and adults, while the present study only focused on primary schoolchildren. In contrast, other countries, including neighbouring Southeast Asian countries to Malaysia, exhibit a relatively lower prevalence of STH infections, ranging from 18.3 to 54.0% (Cho et al., 2021; Dawaki et al., 2019; Kurscheid et al., 2020; Molla & Mamo, 2018; Moncayo et al., 2018; Wijaya et al., 2021). This variation in prevalence may arise from disparities in sample size, sampled population, geographical regions, and SES.

Based on stratification by regions and schools, a significantly higher prevalence of STH infection was seen in the schools in Peninsular Malaysia compared to those in Borneo Malaysia. The highest prevalence was observed in Perak (96.6%), followed by Pahang (85.4%), Johor (42.1%) and Sabah (6.2%), while none of the schoolchildren in Sarawak exhibited STH infection. Various factors may contribute to the STH infection variations observed in different regions and schools, including ethnicities, geographical aspects, SES, personal hygiene, behaviour and health status (Aemiro et al.,

2022; Nasr et al., 2020; Yaro et al., 2021). It was noticed that both schools in Sarawak displayed a complete absence of STH infections, which may be due to improved behavioural and sanitation practices. Furthermore, different soil types and climates in these schools may play a role. Certain soil types, such as limestone and sandstone, were unfavourable for helminth proliferation. Generally, clay soils are conducive for *A. lumbricoides* and *T. trichiura* development, while hookworms favour sandy soil for the proliferation of their larvae (Yaro et al., 2021). It is postulated that the hot and arid coastal environment in Sarawak schools may not be conducive to the development of STHs, as these parasites necessitate optimal warmth, moisture, and adequate shade to protect from ultraviolet (UV) for survival and maturation (Aemiro et al., 2022; Knopp et al., 2008). In addition, high salt (sodium chloride) levels in sandy soils near the sea render them unsuitable for certain parasitic species, as they impede development and facilitate easy transportation by seawater, especially in tide line areas along the coast (Ramos et al., 2020).

In this study, the prevalence of *T. trichiura* was the most prevalent, followed by *A. lumbricoides* and hookworm. This finding is in agreement with other studies showing more children being infected by *T. trichiura* in contrast to *A. lumbricoides* and hookworm infections (Ahmad et al., 2014; Ahmed et al., 2012b; Aini et al., 2007; Kaminsky et al., 2014; Kan & Poon, 1987; Lee et al., 2014; Lepper et al., 2018; Mekonnen et al., 2020; Mohd-Shaharuddin et al., 2018; Moncayo et al., 2018; Muslim et al., 2019; Ngui et al., 2015; Papier et al., 2014; Sungkar et al., 2019; Worrell et al., 2016). The high prevalence of *T. trichiura* infection was potentially due to deworming medication resistance, inefficacious dosage and type of medication used. Additionally, it is known that *T. trichiura* infection remains resistant even after the treatment with a triple dosage of deworming medication, which may contribute to the high infection and reinfection rate among these rural schoolchildren (Sungkar et al., 2019). In contrast, other studies reported that *A. lumbricoides* had the highest infection rate when compared to *T. trichiura* and hookworm species (Aemiro et al., 2022; Lee et al., 2014; Nasr et al., 2020; Silver et al., 2018; Widjana & Sutisna, 2000).

Among schoolchildren from schools with a high prevalence of STH infection ($\geq 70\%$), the older age group (10–11 years old) was found to have a higher prevalence of STH infection and *T. trichiura*. Besides, it is one of the significant risk factors for STH infections, based on multivariate logistic regression analysis. This agrees with other studies, which could be explained by the increased exposure of the older age group especially to outdoor activities (Aemiro et al., 2022; Djuardi et al., 2021; Nasr et al., 2020). Our study demonstrated that being female was one of the significant predictors of STH infections. This finding agreed with a study in Indonesia, whereby the prevalence of STH infections was more prevalent among female schoolchildren than male schoolchildren (Darlan et al., 2017). On the other hand, other studies reported no significant relationship between sex and STH infections (Agustaria et al., 2019; Molla & Mamo, 2018). Low monthly household income was also observed to be significantly associated with STH infections. This could be explained by the poor toilet facilities, unclean environment and inadequate sanitation that are generally linked to low SES status.

Furthermore, subpar living conditions, environmental sanitation and hygiene behaviours, such as untreated water supply, absence of toilets (among schools with low prevalence of STH infection), absence of flush toilets (among schools with high prevalence of STH infection), indiscriminate defaecation, indiscriminate garbage disposal, not wearing shoes (among schools with low prevalence of STH infection) and eating with hands were found to be significantly associated to STH infections. Many studies have shown a significant relationship between poor hygiene practices and STH infections (Aemiro et al., 2022; Ngui et al., 2015; Noradilah et al., 2022). In the present study, many children, especially schoolchildren from schools with a high prevalence of STH infection, were still practising

indiscriminate defecation, which may lead to contamination of soils and rivers. Transmission of infections may happen especially near these contaminated areas since many of the rural communities may still rely on untreated water sources such as rivers for consumption and daily domestic use. A prior study reported that these practices of indiscriminate defecation and garbage disposal are common among OA communities, indicating a correspondingly high prevalence of STH infections in this population (Noradilah et al., 2022). In addition, those who practise open defecation are likely to have poor hygiene and sanitation practices and may not practise proper handwashing afterwards. While eating with hands is an acceptable practice, especially in Malaysian culture, eating with dirty hands might increase the risk of contracting STH infections. The high infection rates of STH, especially *A. lumbricoides* and *T. trichiura* infections in our study population, can be explained by the similar transmission mode whereby they infect humans via the faecal-oral route (Jourdan et al., 2018). A study showed that those who did not wash their hands regularly were triple times more likely to have STH infections (Aemiro et al., 2022). A study conducted by Dawaki et al. (2019) also showed a similar behavioural risk factor related to STH infections among rural communities in Nigeria, specifically in not wearing shoes, which contributed to four times the risk of getting STH infections.

To date, this is the first nationwide school-based study that has reported the prevalence of STH infections and the associated risk factors among rural primary schools in Malaysia. Hence, the present study provides more holistic and updated findings on the STH infection status of Malaysian children from rural areas. Based on the findings of the current study and school visit observations, it is suggested that two types of interventions are implemented: behavioural (lifestyle interventions) and health-related (clinical interventions). Behavioural or lifestyle intervention through education and emphasis on good hygiene practices are some of the ways to reduce STH infections among this study's population. A study by Nasr et al. (2013) revealed a lack of information, behaviour and practices on STH among OA communities, while Chew et al. (2022) reported that a lack of health education and language barriers may add to the issue of high undernutrition cases among them. Therefore, adding information regarding STH infections to the primary school syllabus may help improve knowledge and increase awareness among schoolchildren. Also, one of the recommended and practical ways to reduce the prevalence of STH infections in this study's population is by implementing a targeted deworming programme, especially in the endemic areas. More than half of the schoolchildren in this study were not dewormed for at least six months to one year before study commencement. This shows that periodic deworming needs to be done among them as they were having a worrying high infection rate of STHs. As this study provided the data related to STH infections by species according to schools and regions and has shown the highest prevalence of infection was by *T. trichiura*, a targeted approach such as periodical deworming through a time with specific doses should be taken, such as by giving anthelmintic medications for three days consecutively to treat *T. trichiura* infection effectively. Nevertheless, this study has a few limitations. It only collected a single faecal sample during sampling. A more accurate estimate of the prevalence of STH infections would have been obtained if there had been multiple samplings throughout the study. The method used to detect the STH infections was qualitative. Hence, it could not quantify the intensity of infections and identify the species of hookworms.

In summary, STH infections remain prevalent among school-aged children in certain rural communities in Malaysia. Poor SES, living conditions, environmental sanitation, and hygiene behaviours among rural schoolchildren are some of the main contributing factors to STH infections. Hence, this calls for a joint effort between stakeholders for targeted control measures focusing on education, awareness, and deworming interventions among affected communities.

ACKNOWLEDGEMENTS

We want to express heartfelt gratitude and appreciation to all the schoolchildren and parents for their voluntary participation in this study. Sincere acknowledgements to the Ministry of Education (Education Planning and Policy Research Division, State Education Department, District Education Office, Headmasters and schoolteachers), Ministry of Health (Family Health Development Division, District Health Office, Government Health Clinic, medical officers, nurses, medical assistants, healthcare assistant), Department of Orang Asli Development, Mr. Saidon Ishak (assist in terms of language and customary guidance related to OA) and Tok Batin for the approvals and collaborative efforts during all phases of this study. This study would not have been possible without assistance and contributions from MPOB, Universiti Malaya (UM) and Universiti Malaysia Sarawak (UNIMAS). This research was supported (in part) by the Intramural Research Program of the NIH, National Institute of Allergy and Infectious Diseases (NIAID).

Conflict of Interest Statement

The author declares that they have no conflict of interest.

Authors' Contributions

RL, YALL, KTT, SCL, KRS, and RN were involved in the study's conceptualisation and design. SNMJ and PYT performed the project administration, including sample and data collection, handling of resources, sample and data analysis, interpretation of results, manuscript writing, and editing. RL, YALL, KTT, SCL, KRS, and RN reviewed and edited the manuscript. All authors read and approved the final version of the manuscript.

Funding

This work was supported by the RMK-11 (Eleventh Malaysia Plan) Grant – PD219/16 (public finding, managed by MPOB). The funder was not involved in the study design, data collection, analysis, and interpretation, the writing of the article, or the decision to submit it for publication.

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